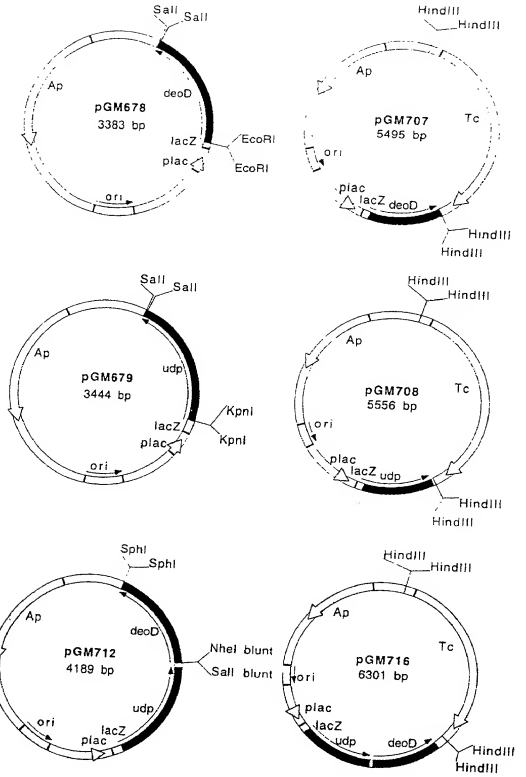

Figure 1.
Cloning vectors for the expression of UdP and PNP enzymes

Plasmid pUC18: 5'sequence of *lacZ gene*

RBS *EcoRI* *KpnI* *SalI* *SphI* *HindIII*

AGGAAAACAGCT ATG ACC ATG ATT ACG AAT TCG AGC TCG GTA CCC GGG GAT CCT CTA GAG TCG ACC TGC AGG CAT GCA AGC TTG

thr met ile thr asn ser ser ser val pro gly asp pro leu glu ser thr cys arg his ala ser leu

plasmid pGM678 and pGM707: sequence of *lacZ-deoD* fused genes

RBS *EcoRI* *SalI*

AGGAAAACAGCT ATG ACC ATG ATT ACG AAT TCT *TCC ATG GCT ACC CCA.....TGG GCG TAA AGAGTAA*GTCGACCTGC....

thr met ile thr asn ser *ser met ala thr pro.....trp ala stop*

plasmid pGM679 and pGM708: sequence of *lacZ-udp* fused genes

RBS *KpnI* *SalI*

AGGAAAACAGCT ATG ACC ATG ATT ACG AAT TCG AGC TCG GTA CCA *TCC ATG TCC .....CTG CTG TAA TTCTCTT*GTCGCAATG....

thr met ile thr asn ser ser ser val *pro ser met ser.....leu leu stop*

palsmid pGM712 e pGM716: 5' and 3' sequence of *deoD gene*

SalI/NheI **RBS** *EcoRI* *SalI* *SphI*

GTCGACTAGCAGGAGGAATTCTTCC *ATG GCT ACC CCA..... TGG GCG TAA AGAGTAA*GTCGACCTGCAGGCATGCAA

met ala thr pro..... trp ala stop

Figure 2. 5′ and 3′ sequences of *udp e deoD* genes cloned in plasmid pUC18. Restriction sites of different constructs are underlined; the ribosome binding site (RBS) is reported in bold. The bases of nucleotide sequence of *udp* and *deoD* genes and the amino acid residues of PNP and UdP proteins are reported in italics.

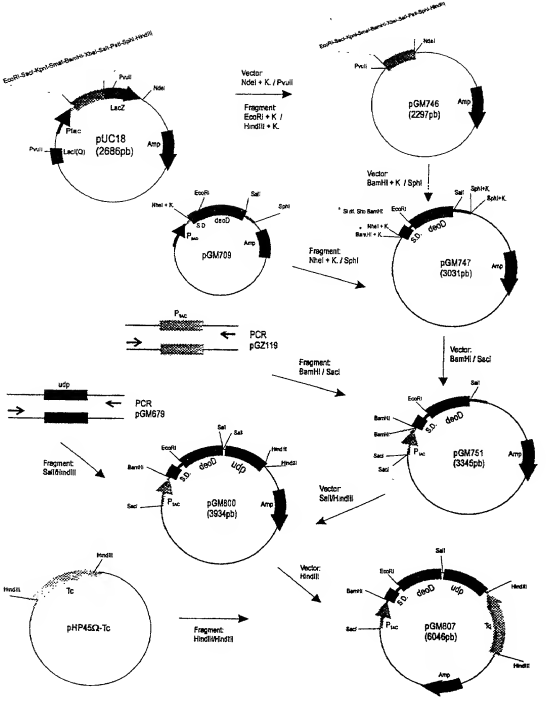

Figure 3.
Costruction of cloning vectors for the expression of UdP and PNP enzymes

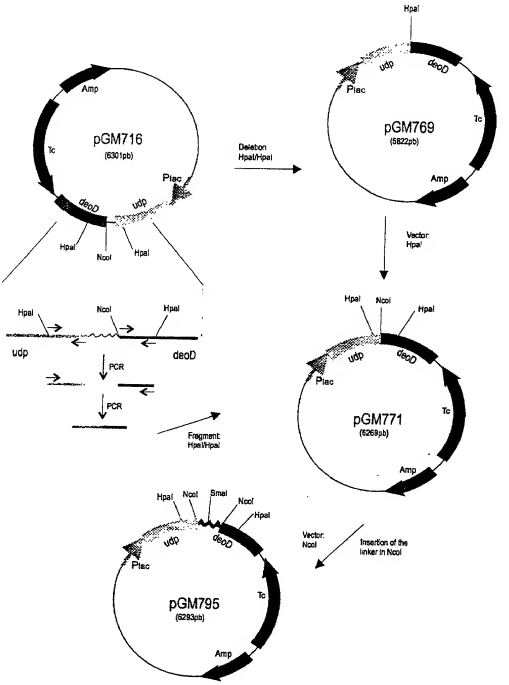

Figure 4.
Construction of cloning vectors for the expression of UdP-(L)-PNP enzymes.

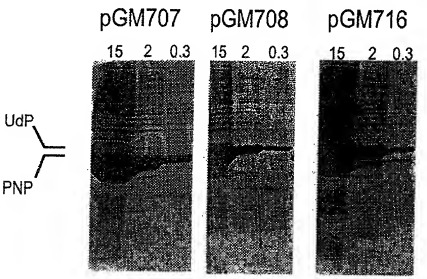

Figure 5.
Expression of PNP and UdP in recombinant *E. Coli* strains. Gel electrophoresis (SDS-PAGE) of total protein extracts from strains MG1655/pGM707, MG1655/pGM708 and MG1655/pGM716 grown over night in LD medium suplemented with 12.5 mg/liter of tetracycline.Lanes 15, 2 and 0.3 correspond to protein extracted from 15, 2 and 0.3 ml of bacterial culture.